

1 **IDENTIFYING PHYSIOLOGICAL TRAITS ASSOCIATED WITH HIGH**
2 **TEMPERATURE TOLERANCE IN SUNFLOWER (*Helianthus annuus* L.)**
3 **GENOTYPES**
4
5
6

7 **ABSTRACT**

8 The objective of the study is to identify the physiological traits associated with seed
9 yield in sunflower. An experiment was conducted at ICAR-Indian Institute of Oilseeds
10 Research, Narkhoda Farm, Rajendranagar, Telangana during *rabi* (Jan-May) 2019 and 2020
11 to find out the response of Sunflower genotypes to high temperature (HT) stress. Here, 47
12 genotypes were screened in the first year and 14 genotypes were identified which are
13 evaluated in the second year under timely (control, S1) and delayed (HT stress, S2) sowings.
14 These traits in the two years are correlated with seed yields (SY) which were differed under
15 S1 and S2. The per se performance indicated that the expression of most traits was reduced
16 under S2 except canopy temperature in 2019. The genotype AKSF 6-3B with the highest SY
17 were characterized by chlorophyll content, maximum quantum efficiency, relative water
18 content, pollen viability, leaf area index, photosynthetic rate and transpiration at vegetative
19 stage under high temperature. The diverse genotypes identified with promising traits can be
20 used in breeding programs to develop new varieties.

21 **KEYWORDS:** correlation, high temperature, seed yield, sunflower, traits.

22 **1. INTRODUCTION:**

23 The climate change drastically affects the agricultural resources (land and water)
24 through increased evapo-transpiration and increased land-degradation, more emission of
25 greenhouse gases, ammonia immobilization, de-nitrification, increasing crop-water
26 requirements, and unavailability of plant-nutrients. The frequency of extreme weather events
27 like heat and cold waves, drought, floods, tropical cyclones and tornadoes, dust and thunder
28 storms, etc., is increasing with negative consequences to the crop productivity.

29 Climate change threatens worldwide crop productivity with new solutions urgently
30 needed to adapt crops to these environmental changes. The domesticated sunflower,
31 *Helianthus annuus*L., is a global oil crop that has promise for adapting to changing
32 environments because it can maintain stable yields across a wide range of environmental
33 conditions, and has been proposed as a potential model crop for adaptation to a changing
34 climate.

35 Climate change effects on crop growth (Yano et al., 2007) development, yield (Lakho
36 et al., 2017), and crop management globally. Even a small fluctuation in temperature can
37 cause more difficulties for crop production. Growth period of many crops was shortened with
38 increasing temperature (Ishaq and Memon, 2016). HT reduces the yield by accelerating
39 phenological phases and by decreasing time of dry matter production (Jan et al., 2017).
40 Hence, climate smart agriculture is a crucial factor to minimize the severe effects of climate
41 change on crop productivity.

42 Sunflower is photo-insensitive crop, grown both in *kharif* and *rabi* predominantly as a
43 rainfed crop. World Sunflower area accounts 27.37 mha, production of 56.07 mt and
44 productivity of 2049 kg ha⁻¹ (Anonymous, 2019). Major countries cultivating sunflower are
45 Russia (8.41 mha), Ukraine (5.95 mha), USA (2.66 mha), Africa (2.21 mha), Argentina (1.88
46 mha), and Romania (1.28 mha) (Anonymous, 2019). The area under this crop in India is 0.26
47 m ha, with a production of 0.22 mt and productivity of 826 kg ha⁻¹. In India, its cultivation is
48 concentrated mainly in states, Karnataka, Maharashtra, Odisha, Andhra Pradesh and Haryana
49 (Anonymous, 2019). 25–30°C is the optimal temperature for germination and growth, while

50 temperatures exceeding 30°C pose stress on the plant. Temperature stress affects plant
51 developmental and physiological processes. Here, the traits involved in HT stress (S2) of
52 different sunflower genotypes and their association with seed yield plant⁻¹ (SY) were
53 identified. The objectives of this study were to: (i) identify the best-performing inbreds under
54 control (S1) and HT (S2), (ii) characterize the association of traits with SY

55 **2. MATERIALS AND METHODS**

56 The experiments were conducted during the late *Rabi* (Jan-May) season of 2019 and 2020
57 at ICAR-IIOR Research Farm, Narkhoda, Hyderabad, India (17°1501600 N, 78°1803000 E;
58 542 m above sea level).

59 *2.1 Plant Material*

60 A set of 47 sunflower (*Helianthus annuus* L.) genotypes including 42 inbred lines and 5
61 hybrid checks were provided by ICAR-IIOR for the experimental study.

62 *2.2 Crop Management and Temperature treatment*

63 The 2019 experiment was carried out using a split-plot design with two temperature
64 treatments using staggered sowings (S1- Timely sowing and S2- Delayed sowing). Each
65 genotype was sown in 0.6×3.6 m plots with a spacing of 60 cm (between rows) x 15 cm
66 (between plants); there were three replicates for each treatment. The response of various
67 physiological traits SPAD chlorophyll readings, membrane thermostability, leaf surface
68 temperature and SY and Heat Susceptibility Index (HSI) to HT were studied and 14
69 genotypes were screened for further study in next year. In depth studies were carried out in late
70 *rabi*, 2020 with the selected sunflower genotypes. The experiment was laid out in split plot
71 design with two temperature treatments i.e., two sowings (timely sowing S1 and delayed
72 sowing S2).

73 *2.3 Physiological Traits*

74 In first season the physiological traits, namely chlorophyll meter readings (SPAD),
75 canopy temperature (CT), and membrane stability index (MSI), were recorded at vegetative
76 and flowering stages using SPAD-502 Plus (Konica Minolta, Inc.), IR thermal gun (AGRI-
77 THERM-6210L; Everest Inter-science Inc.) and Electric Conductivity meter. The canopy
78 temperature measurements were made on sunny days between 10:00 and 13:00 h. The second
79 season evaluation data on the physiological traits, namely relative water content (RWC),
80 photosynthetic rate (PN), stomatal conductance (gS), transpiration rate (E), internal CO₂
81 concentration (Ci), pollen viability, leaf area index (LAI), leaf angle (LA), leaf chlorophyll
82 content (Chl) and maximum quantum yield (Fv/Fm). SY was recorded during both the years.

83 *2.4 Statistical Analysis*

84 The data were summarized using descriptive statistics and analyzed using correlation analysis
85 and CA. Analysis of variance (ANOVA) was conducted for each trait under S1 and S2
86 conditions as described by Panse and Sukhatme (1964). Phenotypic correlations were
87 determined following Johnson et al. (1955).

88 **3 RESULTS**

89 *3.1. Temperature difference during crop growth period*

90 During first year study, the mean maximum (Tmax) and mean minimum temperature
91 (Tmin) from sowing to flowering for S1 were 34.9°C and 17.8°C while for S2 was 37.9°C and
92 20.2°C, from flowering to harvest were 39.4°C and 21.9°C and 41.1°C and 25.0°C
93 respectively. The difference in Tmax recorded in two sowings was 3.0°C at sowing to
94 flowering and 1.7°C at flowering to harvest. During second year study, the Tmax and Tmin
95 from sowing to flowering for S1 was 32.2°C and 16.4°C while for S2 was 35.3°C and 20.2°C.
96 The Tmax and Tmin from flowering to harvest for S1 was 36.1°C and 20.8°C while for S2 was
97 38.3°C and 23.8°C. The difference in Tmax recorded in two sowings was 3.1°C at sowing to
98 flowering and 2.2°C at flowering to harvest.

99 3.2. Mean performance of sunflower genotypes under normal temperature and HT conditions
100 during late rabi, 2019

101 The sunflower genotypes showed variation among most of the traits under S1 as well
102 as S2 conditions (Tables 1). ANOVA indicated that there were significant differences among
103 temperature treatments (A), genotypes (B) and their interaction (A×B). During 2019, the
104 physiological parameters like SPAD value and MSI has reduced under HT.

105 3.2.1. SPAD values

106 Significant variation was observed among the genotypes within the sowing dates for
107 SPAD values during both the vegetative and flowering stages. At vegetative stage, the SPAD
108 values varied significantly from 32.30 to 48.73 with mean 39.92 under S1 and from 21.76 to
109 46.10 with an average 33.78 under S2. The maximum SPAD values were noticed in hybrid
110 check DRSH 1 during S1 (48.73) and S2 (46.10), while inbreds CMS 144B (46.73) under S1
111 and ARM 248B (40.07) under S2 recorded maximum SPAD value. Inbreds CMS 108B (2%),
112 -275B (2%), FMS 400B (2%), ARM 248B (3%) has recorded lowest percent reduction in
113 SPAD reading compared to checks. At flowering SPAD values varied from 30.13 to 45.87
114 under S1 (37.82) and from 19.63 to 42.03 under S2 (31.24). The maximum SPAD values
115 were noticed in hybrid checks KBSH 44 (45.87) under S1 and DRSH 1 (42.03) under S2,
116 while inbred CMS 144B recorded maximum SPAD values under S1 (45.10) and S2 (37.27).
117 Inbreds FMS 400B (0%), CMS 108B (2%) and -275B (3%) has recorded lowest percent
118 reduction in SPAD readings compared to checks.

119 3.2.2. Membrane stability index (MSI)

120 The MSI varied significantly from 25.7 to 45.8 under S1 (36.2) and from 9 to 31.2
121 under S2 (22.1). The maximum MSI values were noticed in inbreds CMS 2023B (45.8%) and
122 NDL 5B (31.2%) under S1 and S2 respectively. The percent reduction in MSI in delayed
123 sowing ranged from 12 to 74 in inbreds and from 15 to 21 in hybrid checks. During
124 flowering, the MSI values varied significantly from 59.5 to 78.6 under S1 (68.6) and from
125 44.3 to 64.1 under S2 (57.4). The maximum MSI values were noticed in inbreds CMS 135B
126 (78.6%) and CMS Pet 2-7-1B (64.1%) under S1 and S2. The percent reduction in MSI in
127 delayed sowing ranged from 7 to 28 in inbreds and from 8 to 15 in hybrid checks. Inbred line
128 AKSF 6-3B has recorded lowest percent reduction in MSI compared to checks during
129 vegetative (12%) and flowering (7%) stages.

130 3.2.3. Canopy temperature

131 During vegetative stage, CT ranged from 22.9°C to 30.4°C under S1 (26.4°C) and
132 from 26.8°C to 34.6°C under S2 (30.7°C). The highest CT was recorded in inbreds CMS
133 275B (30.4°C) and HA 292B (34.6°C) under S1 and S2 respectively. Inbreds AKSF 6-3B
134 (7%), CMS lines-144B (9%), -234B (10%) and -275B (10%) has recorded lowest percent
135 change compared to checks. At flowering, CT varied from 27.0°C to 32.9°C under S1
136 (30.4°C) and from 30.8°C to 35.9°C under S2 (33.6°C). The inbred CMS lines -853B and -
137 2023B recorded maximum CT under S1 and S2. CMS lines-144B (2%), -108B (5%), -275B
138 (5%) has recorded lowest percent change compared to checks.

139 3.2.4. Seed yield (g/plant)

140 There is significant difference for SY between the two sowing dates, in response of
141 genotypic differences within a sowing and between the sowings. Among the genotypes
142 tested, maximum SY was recorded in checks RSFH 130 (32.2), CSFH 12205 (24.3), KBSH
143 44 (21), DRSH 1 (20.1) followed by inbreds NDL 3B (12.4), CMS 519B (10.5) in control
144 (S1). Under S1, the SY ranged from 1.5 to 32.2 whereas in S2 it ranged from 0.1 to 16.5.
145 Maximum SY was recorded among checks KBSH 44 (16.5), CSFH 12205 (10.8), RSFH 130
146 (9.8), DRSH 1 (9.5) followed by inbreds CMS 853B (8.3), -127B (7.3) and AKSF 6-3B (7.1)
147 under HT (S2). Subjecting the plants to a HT resulted in reduction in SY from a mean of 7.5
148 (S1) to 3.9(S2) which is about 13 to 97% reduction in inbreds and 16 to 70% reduction in

149 hybrid checks over control. Inbreds AKSF 6-3B (1%), CMS lines -59B (8%), -127B (10%), -
 150 302B (11%), -135B (12%), -107B (13%) has shown lowest reduction percent compared to
 151 checks.

152 **Table 1:** SPAD values, Membrane stability index (MSI), canopy temperature (CT) and seed
 153 yield (SY) of sunflower during vegetative and flowering stages, *late rabi* 2019

	SPAD value		MSI		CT		SY
	Vegetative stage	Flowering	Vegetative stage	Flowering	Vegetative stage	Flowering	
Mean	36.85	34.53	29.15	63	28.55	32	5.7
Temperature							
CD (P<0.05)	1.92	1.25	1.7	3.3	0	0	0.5
CV (%)	10.2	7.1	11.6	10.1	1	2	16.2
Genotypes							
CD (P<0.05)	4.36	3.19	5	6	1	1	1.3
CV (%)	10.2	8.1	15.2	8.3	4	2	20.2
Interaction	NS	4.59	7.1	NS	2	1	1.9

154 NS-nonsignificant at p<0.05

155 3.2.5. Heat Susceptibility Index

156 The HSI values for the inbreds ranged between -0.28 (CMS 144B) to 2.04 (HA 248B)
 157 and for checks ranged between 0.34 (CO 2) to 1.46 (RSFH 130). CMS lines-144B (-0.28), -
 158 42B (-0.29), -59B (0.14), -127B (0.20), -135B (0.23), -107B (0.27) and AKSF 6-3B (0.03),
 159 recorded lower HSI. Higher values were noted in HA 248B (2.04), CMS 607B (1.81), -103B
 160 (1.76), -850B (1.60) and NDL 3B (1.52). The genotypes with lower HSI values are
 161 considered as heat tolerant. The performance of the inbreds and checks were assessed based
 162 on the field tolerance in the first year and 10 inbreds (6 tolerant and 4 susceptible inbreds)
 163 were selected for the second year study. AKSF 6-3B, CMS lines -42B, -107B, -127B, -135B,
 164 -144B were selected as tolerant genotypes and CMS lines-17B, -70B, -125B, ARM 243B as
 165 susceptible genotypes. CO 2, CSFH 12205, DRSH 1 and KBSH 44 were taken as checks
 166 respectively.

167 3.3. Mean performance of sunflower genotypes under normal temperature and HT conditions 168 during late rabi, 2020

169 3.3.1. Relative Water Content (RWC)

170 During vegetative stage the RWC ranged from 57.9 to 67.3% under S1 (62.7%) and
 171 from 49.2 to 59.4% under S2 (54.4%). Check DRSH 1 under S1 (67.3%) and S2 (59.4%) has
 172 highest RWC (Table 2). At flowering, RWC varied from 63.8 to 68.0% under S1 (63.8%) and
 173 from 51.1 to 61.0% under S2 (55.6%). Checks DRSH 1, KBSH 44 noted maximum RWC
 174 under both S1 and S2. Inbred AKSF 6-3B at vegetative (4%) and flowering (2%) has
 175 recorded least percent reduction for the trait RWC compared to checks.

176 3.4. Photosynthetic parameters

177 3.4.1. Photosynthetic Rate (P_N) $\mu\text{mol}(\text{CO}_2) \text{m}^{-2} \text{s}^{-1}$

178 Significant negative effect due to HT was observed among the genotypes for P_N
 179 during both vegetative and reproductive stages. At vegetative stage P_N varied from 16.74 to
 180 29.88 with a mean of 23.6 in S1 and from 12.6 to 26.36 with a mean of 19.9 in S2. In S1
 181 check CSFH 12205 (29.88), followed by inbred AKSF 6-3B (27.99) and in S2 condition
 182 inbred AKSF 6-3B (26.36), followed by check CSFH 12205 (25.78), recorded highest P_N
 183 values. Inbred AKSF 6-3B (6%) has recorded least percent reduction compared to checks. At
 184 flowering P_N varied from 17.6 to 34.4 with a mean of 25.6 in S1 and from 13.3 to 32.8 with a
 185 mean of 22.4 in S2. Inbred AKSF 6-3B, followed by checks CSFH 12205, DRSH 1 has
 186 recorded maximum P_N . Inbred AKSF 6-3B has recorded least percent reduction in P_N at
 187 vegetative (6%) and flowering (0%) compared to checks.

188 3.4.2. Stomatal conductance (g_s) $\text{mol H}_2\text{O m}^{-2} \text{s}^{-1}$

189 There is significant difference for g_s were observed between the two sowing dates
 190 during both vegetative and flowering stages. During vegetative stage, g_s varied from 90 to
 191 145 under S1 (110) and from 60 to 109 under S2 (81). At flowering, g_s varied from 94 to 150

192 (119) in S1 and from 64 to 113 (89) in S2 condition. During both stages, CMS 135B and
193 AKSF 6-3B recorded maximum gs under S1 and S2 respectively. Inbred AKSF 6-3B has
194 recorded least percent reduction in gs at vegetative (3%) and flowering (6%) compared to
195 checks.

196 3.4.3. Transpiration (E)

197 Genotypes differed significantly for E with the sowing dates during both vegetative
198 and flowering stage. During vegetative stage, E value varied from 1.1 to 2.9(2)in S1 and from
199 0.9 to 2.1(1.6) in S2. Inbreds CMS 42B (2.9), -135B (2.4), followed by check KBSH 44 (2.3)
200 in S1 while checks CO 2 (2.1), KBSH 44 (2) has highest E values in S2. At HT a reduction in
201 E values was observed among all the genotypes. At flowering, E ranged from 2.1 to 3.8(3) in
202 S1 and from 1.2 to 3.1(2.2) in S2. Inbreds CMS 135B (3.8), -70B (3.6) in S1 while check
203 DRSH 1 (2.6), KBSH 44 (2.6), and inbred ARM 243B (2.6) in S2 has highest E. Inbred
204 AKSF 6-3B has recorded least percent reduction in E at vegetative (5%) and flowering (10%)
205 stages compared to checks.

206 3.4.4. Internal CO₂ concentration (C_i)ppm

207 C_i at vegetative stage varied from 447 to 711(571) in S1 and from 364 to 624 (465)in
208 S2. In S1, inbred CMS 125B (711), followed by check DRSH 1 (705) and in S2, check
209 DRSH 1 (624), followed by inbred ARM 243B (537) has highest C_i. Inbreds AKSF 6-3B
210 (11%), ARM 243B (16%) and CMS 107B (17%) has recorded percent reduction on par with
211 checks. At flowering, C_i varied from 490 to 793 (627) in S1 and from 379 to 631 (503) in S2.
212 In S1 inbreds CMS 125B (793), followed by CMS 127B (757), check DRSH 1 (721), and in
213 S2, check DRSH 1 (631), followed by inbred CMS 127B (589), has recorded maximum C_i.
214 Inbred AKSF 6-3B (10%) has recorded percent reduction on par with checks.

215 Li et al. (2019) observed HT treatment resulted in a significant increase in stomatal
216 conductance and transpiration rate in rice. Khetrapal et al. (2009) reported that stomatal
217 conductance of chickpea crop decreased under elevated temperature significantly at all
218 growth stages. Balla et al. (2019) observed increased evaporation occurred in spite of the
219 strong reductions in stomatal conductance, unrelated to the phenophase.

220 3.4.5. Pollen Viability (PV)

221 PV ranged from 76 to 91.9 (85.4) in S1 and from 58 to 88.7 (74.8) in S2. PV was
222 reduced by 3 to 7% among checks and by 1 to 29% among inbreds at HT over control.
223 Among the genotypes maximum reduction in PV was observed in inbred CMS lines -125B
224 (29%), -135B (23%), -42B (21%) and minimum reduction was observed in AKSF 6-3B (1%).
225 Kaur and Behl (2010) reported HT of above 30°C during floret development may cause
226 complete sterility in wheat depending on genotypes.

227 3.4.6. Leaf Area Index (LAI)

228 During vegetative and flowering stages significant differences for LAI were observed
229 among the genotypes, with the two sowing dates and the interaction effect. During vegetative
230 stage LAI ranged from 0.72 to 1.44 (1.02) in S1 and from 0.46 to 1.14 (0.78) in S2.
231 Maximum LAI was observed in check KBSH 44 (1.44), followed by inbreds ARM 243B
232 (1.42), AKSF 6-3B (1.21) in S1 and in check KBSH 44 (1.14), followed by inbreds AKSF 6-
233 3B (0.95), CMS 42B (0.93) in S2. The percent reduction in LAI due to delayed sowing
234 ranged from 12 to 68 in inbreds and from 9 to 21 in checks. Inbreds CMS 127B (12%), -107B
235 (13%), -42B (14%), -144B (17%), -70B (20%) has recorded percent reduction on par with
236 checks. At flowering, LAI ranged from 1.01 to 4.04 (2.57) in S1 and from 0.69 to 3.86 (2.31)
237 in S2. Inbred AKSF 6-3B (3%) has recorded least percent reduction compared to checks.

238 3.4.7. Leaf Angle (LA)

239 At vegetative stage, LA ranged from 39 to 58°(45°)in S1 and from 29 to 50°(38°) in
240 S2. Maximum LA was recorded in inbreds CMS 42B followed by check CO 2 (48°) in S1
241 and S2. Minimum LA was noted in inbreds AKSF 6-3B (39°), CMS 107B (39°) and CMS

242 127B (39°) in S1 and in inbreds CMS 125B (29°), -17B (32°) and -127B (32°) in S2. The
 243 percent reduction in LA due to delayed sowing varied from 3 to 8 among checks and from 5
 244 to 36 among the inbreds. At flowering stage, LA ranged from 33 to 52° in S1 (39°) and from
 245 23 to 46° in S2 (33°). Maximum LA was recorded in inbred CMS 42B followed by check
 246 DRSH 1 in both S1 and S2. Minimum LA was recorded in inbreds AKSF 6-3B (33°), CMS
 247 125B (33°), -17B (36°) and ARM 243B (36°) in S1 and in inbreds CMS 125B (23°), -17B
 248 (28°) and in AKSF 6-3B (30°) in S2. The percent reduction in leaf angle due to delayed
 249 sowing varied from 7 to 13 among checks and from 8 to 30 among the inbreds.

250 3.4.8. Total Chlorophyll(TC) mg g⁻¹ FW

251 In vegetative stage, the TC varied from 2.20 to 4.30 (3.60) in S1 and from 1.46 to
 252 3.98 (3.11) in S2. Among the genotypes maximum TC was recorded in checks DRSH 1,
 253 CSFH 12205, KBSH 44, CO 2 in S1 and S2 conditions respectively. The percent reduction in
 254 TC due to delayed sowing (HT) ranged from 4 to 34 in inbreds and from 6 to 10 in checks. At
 255 flowering stage TC varied from 3.55 to 6.04 (4.88) in S1 and from 2.36 to 5.36 (4.01) in S2.
 256 Maximum TC was noted in checks DRSH 1, KBSH 44, CO 2, CSFH 12205 among both the
 257 sowings. The percent reduction in TC due to delayed sowing (HT) ranged from 9 to 34 in
 258 inbreds and from 8 to 14 in checks. Inbred AKSF 6-3B has recorded percent reduction on par
 259 with checks during both vegetative (4%) and flowering stages (9%). Mohammed and Tarpley
 260 (2010) reported a decline in Chl pigment as a result of lipid peroxidation of chloroplast and
 261 thylakoid membranes in sorghum due to heat stress (40/30°C, day/night).

262 **Table 2:** Physiological parameters and seed yield of sunflower genotypes in vegetative (veg)
 263 and flowering (flow) stages during *late rabi*, 2020

	RWC		P _N μmol (CO ₂) m ⁻² s ⁻¹		g _s mol (H ₂ O) m ⁻² s ⁻¹		E mmol (H ₂ O) m ⁻² s ⁻¹		C _i ppm		PV
	Vegetative	Flowering	Vegetative	Flowering	Vegetative	Flowering	Vegetative	Flowering	Vegetative	Flowering	
Mean	58.5	59.7	21.6	24	95.5	104	1.8	2.6	518	567.5	80.1
Mainplot Temperature treatments											
CD (P<0.05)	1.3	1	3.3	2.9	18	5	0.2	0.2	66	105	1.5
CV (%)	2.4	1.8	16.2	12.7	20	5	13.7	8.8	14	20	2.1
Subplot Genotypes											
CD (P<0.05)	NS	NS	3.9	2.1	NS	20	0.3	0.6	107	77	4
CV (%)	7.4	7.5	15.7	7.4	23	17	15.9	8.8	18	12	4.3
Interaction	NS	NS	NS	NS	NS	29	NS	NS	NS	NS	5.7

264

	LAI		LA		TC		Fv/Fm		SY
	Vegetative	Flowering	Vegetative	Flowering	Vegetative	Flowering	Vegetative	Flowering	
Mean	0.9	2.44	41.5	36	3.355	4.445	0.727	0.7515	16.55
Mainplot Temperature treatments									
CD (P<0.05)	0.08	NS	7	2	0.3	0.36	0.04	NS	1.7
CV (%)	8.9	13.4	17	5	3.97	8.53	5.8	17.1	11.2
Subplot Genotypes									
CD (P<0.05)	0.12	0.25	7	7	0.38	0.47	0.07	NS	1.4
CV (%)	11.9	8.8	13	16	9.8	9.22	8.3	8.5	23.2
Interaction	0.18	NS	NS	NS	NS	NS	NS	NS	NS

265 RWC relative water content, P_N photosynthetic rate, g_s stomatal conductance, E transpiration, C_i internal CO₂ concentration,
 266 LAI leaf area index, LA leaf angle, TC total chlorophyll, Fv/Fm maximum quantum efficiency, SY seed yield.

267 3.4.9. Maximum Quantum efficiency (Fv/Fm)

268 During vegetative stage, significant differences for Fv/Fm were observed with the two
 269 sowing dates among the genotypes. In S1, inbreds AKSF 6-3B (0.816), CMS 107B (0.814),
 270 followed by checks CO 2 (0.807), KBSH 44 (0.807) and in S2, check CO 2 (0.786), inbred
 271 AKSF 6-3B (0.772), checks DRSH 1 (0.732), KBSH 44 (0.720) has maximum Fv/Fm. The
 272 percent reduction in Fv/Fm due to HT ranged from 5 to 26 in inbreds and from 3 to 13 in
 273 checks. Inbreds AKSF 6-3B (5%) and ARM 243B (13%) recorded on par reduction percent
 274 for the trait Fv/Fm during vegetative stage compared to checks. At flowering, maximum
 275 Fv/Fm noted in inbred AKSF 6-3B (0.837) followed by check CO 2 (0.830), inbred CMS
 276 107B (0.821) in S1 and in inbred AKSF 6-3B (0.776), followed by checks CO 2 (0.752),
 277 DRSH 1 (0.738) in S2. The percent reduction in Fv/Fm due to HT ranged from 7 to 21 in

278 inbreds and from 9 to 10 in checks. Inbred AKSF 6-3B (7%) recorded least reduction percent
279 for the trait Fv/Fm during flowering stage compared to checks.

280 3.4.10. Seed yield (SY) g/pl

281 Genotypic difference was significant for SY with the sowing dates. Subjecting the
282 plants to a HT resulted in reduction in SY from a mean of 18.5 (S1) to 14.6 (S2) which is
283 about 3 to 10% reduction among checks and 3 to 47% reduction among the inbreds over
284 control. Inbred line AKSF 6-3B (3%) has recorded least percent reduction among the inbreds
285 and on par with checks. Inbred CMS lines -17B and -107B showed on par SY with checks
286 CO-2 and CSFH-12205. Among the genotypes tested, maximum SY was recorded in checks
287 KBSH 44 (25.3), DRSH 1 (25.1), followed by CMS 17B (21.6) and AKSF 6-3B (20.8) in S1
288 and in checks DRSH 1 (23.7), KBSH 44 (23.5), followed by AKSF 6-3B (20.5) in S2.

289 3.4.12. Association of traits with SY

290 The traits associated with SY can be used to aid the selection of superior genotypes.
291 Under first year, the correlation analysis revealed that the traits SPAD value at both the
292 stages, were positively correlated with SY under S1. The traits SPAD value and MSI at both
293 vegetative and flowering stages were positively correlated with SY while CT at both
294 vegetative and flowering stages were negatively correlated with SY under S2.

295 Under second year, the traits like PV were positively correlated with SY under S1.
296 The traits like TChl, Fv/Fm, RWC, LAI at vegetative and flowering stages, PN, transpiration
297 at vegetative stage, PV were positively correlated with SY under S2. CT was negatively
298 correlated with SY, indicating that higher CTs limit the yield of genotypes.

299 3.5 DISCUSSION

300 Chlorophyll synthesis is sensitive to HS and is a good indicator of HS injury (Gosavi
301 et al., 2014). Lower SPAD meter value indicates the decreased P_N and the inability of a plant
302 to produce higher yield.

303 Cell membrane stability varies with the age of plant tissue, growth stage, growing
304 season, plant species, and intensity of heat stress (Nijabat et al., 2020). HS inactivates the
305 enzymes and denatures the membrane protein, resulting in membrane permeability and
306 integrity changes causing reduction in ion flux, electrolyte leakage, production of toxic
307 compounds, changes in RWC, and disruption of homeostasis thus reducing cell viability (Hu
308 et al., 2020). HT leads to swelling of grana stacks and an aberrant stacking, accompanied by
309 ion-leakage from leaf cells and changes in energy allocation to the photosystems
310 (Allakhverdiev et al., 2008). The maintenance of cellular membrane function under HS is
311 essential for a sustained photosynthetic performance (Chen et al., 2010).

312 CT is the more accurate estimate of the consequences of HS on the crop than air
313 temperature (Gabaldon-Leal et al., 2016). Small lobed leaves and conifers tend to reach lower
314 temperatures whereas, large leaves tend to reach higher temperatures (Leuzinger et al. 2010).
315 An increase in average air temperature results in higher mean canopy temperature during
316 anthesis.

317 The leaf RWC is the result of the equilibrium between water absorption and evapo
318 transpiration (Rodriguez et al., 2005). At HT, water absorption through the roots is promoted.
319 The movement of water within the plant is attributed to changes in membrane fluidity and
320 permeability, changes in water viscosity or a combination of both (Iglesias-Acosta et al,
321 2010). HS affects plant root conductance despite of enough water supply and this becomes
322 more fatal when HS is combined with drought (Sattar et al., 2020).

323 In chloroplasts, carbon metabolism of the stroma and photochemical reactions in
324 thylakoid lamellae are the primary sites of injury at HT (Wang et al., 2008). HT reduce the
325 activation state of ribulose-1,5-bisphosphate carboxylase/ oxygenase (rubisco) (Han et al.,
326 2009), which has often been ascribed to thermolability and the loss of activity in rubisco
327 activity (Sharkey, 2010) or by changing the affinity of RubisCO for CO_2 (Berry 1980). By

328 increasing chlorophyllase activity and decreasing the amount of photosynthetic pigments, HS
329 ultimately reduces the plant photosynthetic and respiratory activity (Todorov et al., 2003).
330 HT disrupts the metabolic processes in the guard cells thus stomatal response is often
331 complicated as temperature affects photosynthesis, VPD, transpiration, and plant water status,
332 which all feedback on stomatal behaviour (Urban et al., 2017). Higher VPD increases the leaf
333 atmosphere diffusion gradient, driving greater water loss (Grashoff et al., 1994) and C_i (Greer
334 and Weedon, 2012) which effect the stomatal closure to maintain plant water status (Mott and
335 Peak, 2013). Evaporative demand as determined by the vapour pressure deficit would
336 increase by about 5 to 6% per degree warming (McKenney and Rosenberg, 1993). HS-
337 induced damage to chloroplasts leads to the inactivation of heat-sensitive proteins such as
338 Rubisco activase (RCA) and the down-regulation of important chloroplast components,
339 thereby leading to decreased photosynthetic efficiency, redox imbalance and possible cell
340 death (Li et al., 2018).

341 HT alters carbohydrate accumulation thus decreasing the availability of energy
342 resources and osmotic power of carbohydrates, leading to a failure in pollen development
343 (Pauplere and Heusden, 2014). These results are supported in sorghum in which the decrease
344 of PV under HS was mainly correlated with a decrease of sucrose and starch in late stages of
345 pollen development, due to decreased expression of several sugar metabolism genes (Jain et
346 al., 2007). HS during pre-anthesis (sporogenesis) decreases PV and fewer pollen grains, in
347 grain sorghum (Prasad et al., 2008), at heading stage significantly reduced anther dehiscence
348 and pollen fertility rate in rice (Ahmed et al., 2010) and at floret development alters pollen
349 morphology and results in an abnormal exine wall, degeneration of tapetum cells, and
350 membrane damage in grain sorghum (Djanaguiraman et al., 2014), wheat (Prasad and
351 Djanaguiraman, 2011) leading to pollen sterility. HS adversely affects pollen cell and
352 microspore resulting into male sterility (Anjum et al., 2008) which reduces the yield due to
353 impairment of pollen development (Hedhly et al., 2011).

354 Reduction in photosynthesis at HT was reflected in reduced LAI. Warmer conditions
355 both accelerate rate of organ initiation and shorten duration of organ growth thereby leading
356 to reduced growth of plant organs (Chakrabarti et al., 2013). Temperature modifies leaf
357 orientation by affecting pulvinal region tissues resulting in active heliotropic movements
358 (Bielenberg et al., 2003). Downward leaf inclination was a general response of the species and
359 is considered to be a susceptible reaction to HS (Kalyar et al., 2013). HT induce differential
360 petiole-driven upward growth of leaves by allocating active resources to the petiole and leaf
361 veins to keep them in a static position (Van Zanten et al., 2009). Plants adjust their canopy
362 according to irradiance while petioles were stiffer at HT (Hernandez, 2010).

363 Under HS, a decrease in chlorophyll biosynthesis due to inhibition of photosynthetic
364 electron transport chain (Mohanty et al., 1989) and the inhibition of enzymes. HT alters the
365 anatomical structure in leaves, and result in reduced photosynthetic and respiratory activities
366 (Lipiec et al., 2013). Therefore, the amount of Chl content strongly depends on the species
367 physiological responses and their ability to tolerate stress. HS results in plant leaf pigment
368 loss and significantly damages photosynthetic activities (Awasthi et al., 2014). The decrease
369 in chlorophyll content was due to the inhibition of chlorophyll biosynthesis or chlorophyll
370 degradation (Dutta et al., 2009). The effect of HT on the pigments and other photosynthetic
371 apparatus is due to the production of toxic oxygen species (oxidative damage) and reduction
372 in antioxidative defense (Kumar et al., 2013). The loss of chlorophyll is typical of leaf
373 senescence and is used as an indicator (Rossi et al., 2017).

374 The approximate optimal Fv/Fm value for many of the crop species is in the range of
375 0.79 to 0.84, with lowered values indicating plant stress (Maxwell et al., 2000). A decrease in
376 the Fv/Fm values indicated a reduction in PSII efficiency, mainly by photoinhibition (Guidi
377 et al., 2019) under HS. Enzyme degradation at HT impede the function of PSII, decrease

378 electron transport rates, inhibit Rubisco activity and decrease chlorophyll content (Prasad and
379 Djanaguiraman, 2011). Use of chlorophyll fluorescence measurements have been shown to
380 be useful in quantifying the impact of drought and heat stress on plants (Ristic et al., 2007).

381 The decrease in SY with late plantings is attributable to higher air temperatures at
382 seed development period, and thus pollination and fertilization were obstructed resulting in
383 hastened maturity, poor seed setting, lesser accumulation/translocation of metabolites. Major
384 yield losses were mainly attributed to limited nutrient translocation rather than a reduction in
385 photosynthetic production under HT and poor quality oil compared to timely sowings. HT
386 affect the SY by affecting phenological development processes as reduction in SN and
387 increase in small grains. Loss of productivity in HS is chiefly related to decreased
388 assimilatory capacity (Hay and Porter, 2006) which is due to reduced photosynthesis by
389 altered membrane stability (Zhang et al., 2006) and enhanced maintenance respiration costs
390 (Reynolds et al., 2007), reduction in radiation use efficiency.

391 Heat indices used for screening heat tolerant genotypes provides a measure of yield
392 loss under HT conditions in comparison to normal as done in case of drought stress by Mitra
393 (2008). Heat susceptibility index (HSI) was calculated to determine the most desirable heat
394 tolerant genotypes

395 **3.6 CONCLUSION**

396 The traits associated with SY under S1 and S2 differed among the sunflower genotypes
397 studied. The inbreds and hybrids with different genetic backgrounds resulted in trait
398 variation. Variation in the S2 condition of specific traits measured among genotypes aids the
399 selection of these traits. These trait-specific genotypes could be used in sunflower breeding
400 programs to develop location-specific varieties. The values of most of the traits were reduced
401 under S2.

402 **3.7 Acknowledgement**

403 The authors like to acknowledge the support, facilities and the plant material provided by
404 ICAR-IIOR (crop production division) for the Ph. D research work and their help during
405 experimental study.

406 There is no conflict of interest.

407 **3.8 References**

408 Allakhverdiev, S.I., Kreslavski, V.D., Klimov, V.V., Los, D.A., Carpentier, R., Mohanty, P.,
409 2008. Heat stress: an overview of molecular responses in photosynthesis. *Photosynthesis*
410 *Research* 98(1), 541–550.

411 Anonymous, Oilseeds data (2019) Directorate of Economics and Statistics. Advance
412 estimate. Five years series data 2014-2019 (Source: <https://eands.dacnet.nic.in>).

413 Awasthi, R., Kaushal, N., Vadez, V., Turner, N.C., Berger, J., Siddique, K.H.M., Nayyar, H.,
414 2014. Individual and combined effects of transient drought and heat stress on carbon
415 assimilation and seed filling in chickpea. *Functional Plant Biology* 41(11), 1148–1167.

416 Balla, K., Karsai, I., Bonis, P., Kiss, T., Berki, Z., Horvath, A., Mayer, M., Bencze, S., Veisz,
417 O., 2019. Heat stress responses in a large set of winter wheat cultivars (*Triticum aestivum* L.)
418 depend on the timing and duration of stress. *PLoS ONE* 14(9), e0222639.

419 Berry J and Bjorkman O. 1980. Photosynthetic response and adaptation to temperature in
420 higher plants. *Annu Rev Plant Physiol* 31:491–543

421 Bielenberg, D.G., Miller, J.D., Berg, V.S., 2003. Paraheliotropism in two Phaseolus species:
422 combined effects of photon flux density and pulvinus temperature, and consequences for leaf
423 gas exchange. *Environmental and Experimental Botany* 49(2), 95–105.

424 Chakrabarti., B., Singh, S.D., Kumar, V., Harit, R.C., Misra, S., 2013. Growth and yield
425 response of wheat and chickpea crops under HT. *Indian Journal of Plant Physiology* 18(1),
426 7–14.

427 Chen, J., Wang, P., Mi, H.L., Chen, G.Y., Xu, D.Q., 2010. Reversible association of ribulose-
428 1,5-bisphosphate carboxylase/oxygenase activase with the thylakoid membrane depends upon
429 the ATP level and pH in rice without heat stress. *Journal of Experimental Botany* 61(11),
430 2939–2950.

431 Djanaguiraman, M., Prasad, P.V.V., Murugan, M., Perumal, M., Reddy, U.K., 2014.
432 Physiological differences among sorghum (*Sorghum bicolor* L.) genotypes under high
433 temperature stress. *Environmental and Experimental Botany* 100, 43–54.
434 <https://doi.org/10.1016/j.envexpbot.2013.11.013>

435 Dutta, S., Mohanty, S., Tripathy, B.C., 2009. Role of Temperature Stress on Chloroplast
436 Biogenesis and Protein Import in Pea. *Plant Physiology* 150(2), 1050–1061.

437 Gabaldón-Leal, C., Webber, H., Otegui, M., Slafer, G., Ordóñez, R., Gaiser, T., Lorite, I.,
438 Ruiz-Ramos, M., Ewert, F., 2016. Modelling the impact of heat stress on maize yield
439 formation, *Field Crop Res*, 198, 226-237. <https://doi.org/10.1016/j.fcr.2016.08.013>

440 Gosavi, G.U., Jadhav, A.S., Kale, A.A., Gadakh, S.R., Pawar, B.D, Chimote, V., 2014. Effect
441 of heat stress on proline, chlorophyll content, heat shock proteins and antioxidant enzyme
442 activity in sorghum (*Sorghum bicolor* L.) at seedlings stage. *Indian Journal of Biotechnology*
443 13(3), 356-363 .

444 Grashoff, C., Rabbinge, R, Nonhebel, S., 1994. Potential effects of global climate change on
445 cool season food legume productivity. In: *Expanding the Production and Use of Cool Season*
446 *Food Legumes*. F.J. Muehlbauer and W.J. Kaiser (eds.). Kluwer Academic, Dordrecht, 159-
447 174. <https://edepot.wur.nl/216092>

448 Greer, D.H, Weedon, M.M., 2012. Modelling photosynthetic responses to temperature of
449 grapevine (*Vitis vinifera* cv: Semillon) leaves on vines grown in a hot climate. *Plant Cell*
450 *Environment*, 35(6), 1050–1064.

451 Guidi, L., Lo Piccolo, E, Landi, M., 2019. Chlorophyll fluorescence, Photoinhibition and
452 Abiotic Stress: Does It Make Any Difference the Fact to Be a C3 or C4 Species? *Front. Plant*
453 *Sci*, 10, p.174.

454 Han F, Chen H, Li X-J, Yang M-F, Liu G-S, Shen S-H. 2009. A comparative proteomic
455 analysis of rice seedlings under various high-temperature stresses. *Biochimica et Biophysica*
456 *Acta*1794: 1625 –1634.

457 Hay, R.K.M., Porter, J.R., 2006. *The physiology of crop yield*. 2nd Edition Blackwell
458 Publishing Ltd; Oxford, UK. <http://dx.doi.org/10.1093/aob/mcm187>

459 Hedhly, A., 2011. Sensitivity of flowering plant gametophytes to temperature fluctuations.
460 *Environmental and Experimental Botany*, 74, 9–16.

461 Hernandez, L.F., 2010. Leaf angle and light interception in sunflower (*Helianthus annuus*
462 L.). Role of the petiole's mechanical and anatomical properties. *Phyton* 79, 109–113.

463 Hu, S., Ding, Y., & Zhu, C. (2020). Sensitivity and responses of chloroplasts to heat stress in
464 plants. *Frontiers in Plant Science*, 11, 375.

465 Iglesias-Acosta, M., Carmen Martinez-Ballesta, M., Antonio Teruel, J., Carvajal, M., 2010.
466 The response of broccoli plants to high temperature and possible role of root aquaporins.
467 *Environmental and Experimental Botany* 68(1), 83–90.

468 Ishaq, W., Memon, S.Q., 2016. Roles of women in agriculture: a case study of rural Lahore,
469 Pakistan. *Journal of Rural Development and Agriculture* 1(1), 1–11

470 Jain, M., Prasad, P.V., Boote, K.J., Hartwell, A.L, Chourey, P.S., 2007. Effects of season-
471 long high temperature growth conditions on sugar-to-starch metabolism in developing
472 microspores of grain sorghum (*Sorghum bicolor*). *Planta* 227(1), 67–79.

473 Jan, S.A., Bibi, N., Shinwari, K.S., Rabbani, M.A., Ullah, S., Qadir, A., Khan, N., 2017.
474 Impact of salt, drought, heat and frost stresses on morpho-biochemical and physiological
475 properties of brassica species: an updated review. *Journal of Rural Development and*
476 *Agriculture* 2, 1–10.

477 Johnson, H.W., Robinson, H., & Comstock, R. 1955. Estimates of genetic and environmental
478 variability in soybeans. *Agronomy Journal*, 47(7): 314– 318.

479 Kalyar, T., Rauf, S., Teixeira da Silva, J.A., Haidar, S., Iqbal, Z., 2013. Utilization of leaf
480 temperature for selection of leaf gas exchange traits for the induction of heat resistance in
481 sunflower (*Helianthus annuus* L.). *Photosynthetica* 51 (3), 419–428.

482 Khetrupal, S., Pal, M., Lata, S., 2009. Effect of elevated temperature on growth and
483 physiological characteristics in chickpea cultivars. *Indian Journal of Plant Physiology* 14(4),
484 377–383.

485 Kumar, S., Thakur, P., Kaushal, N., Malik, J.A., Gaur, P, Nayyar, H., 2013. Effect of varying
486 high temperatures during reproductive growth on reproductive function, oxidative stress
487 and seed yield in chickpea genotypes differing in heat sensitivity. *Archives of Agronomy and*
488 *Soil Science* 59, 823–843. <http://dx.doi.org/10.1080/03650340.2012.683424>

489 Leuzinger, S., Vogt, R., Korner, C., 2010. Tree surface temperature in an urban environment.
490 *Agricultural and Forest Meteorology* 150(1), 56–62.

491 Li, X., Cai, C., Wang, Z., Fan, B., Zhu, C., Chen, Z., 2018. Plastid translation elongation
492 factor Tu is prone to heat-induced aggregation despite its critical role in plant heat tolerance.
493 *Plant Physiology* 176(4), 3027–3045.

494 Lipiec, J., Doussan, C., Nosalewicz, A., Kondracka, K., 2013. Effect of drought and heat
495 stresses on plant growth and yield: a review. *International Agrophysics* 27(4), 463–477.

496 Maxwell, K., Johnson, G.N., 2000. Chlorophyll fluorescence-A practical guide. *Journal of*
497 *Experimental Botany* 51(345), 659–668.

498 McKenney, M.S., Rosenberg, N.J., 1993. Sensitivity of some potential evapotranspiration
499 estimation methods to climate change. *Agricultural and Forest. Meteorology* 64(1), 81-110.

500 Mitra, R., Bhatia, C.R., 2008. Bioenergetic cost of heat tolerance in wheat crop. *Current*
501 *Science* 94(8): 1049–1053.

502 Mohanty, N., Vass, I., Demeter, S., 1989. Impairment of photosystem 2 activity at the level
503 of secondary quinone acceptor in chloroplasts treated with cobalt, nickel and zinc ions
504 *Physiologia Plantarum* 76(3), 386–390.

505 Mohammed, A.R., Tarpley, L., 2010. Effects of high night temperature and spikelet position
506 on yield-related parameters of rice (*Oryza sativa* L.) plants. *European Journal of Agronomy*
507 33(2), 117-123.

508 Mott, K.A., Peak, D., 2010. Stomatal responses to humidity and temperature in darkness.
509 *Plant, Cell and Environment* 33(7), 1084–1090.

510 Nijabat, A., Bolton, A., Mahmood-ur-Rehman, M., Shah, A.I., Hussain, R., Naveed, N.H.,
511 Ali, A., Simon, P., 2020. Cell membrane stability and relative cell injury in response to heat
512 stress during early and late seedling stages of diverse carrot (*Daucus carota* L.) germplasm,
513 *Horticulture Science* 55(9), 1446-1452.

514 Panse, V.G and Sukhatme, P. V. 1967. “Statistical Methods for Agricultural Workers,” 2nd
515 Edition, Indian Council of Agricultural Research, New Delhi.

516 Paupiere, M.J, van Heusden, A.W, Bovy, A.G. 2014. The metabolic basis of pollen thermo-
517 tolerance: perspectives for breeding. *Metabolites*, 30 (4):889-920.

518 Prasad, P.V.V., Djanaguiraman, M., 2011. High night temperature decreases leaf
519 photosynthesis and pollen function in grain sorghum. *Functional Plant Biology* 38(12), 993–
520 1003.

521 Prasad, P.V.V., Pisipati, S.R., Mutava, R.N., Tuinstra, M.R., 2008. Sensitivity of grain
522 sorghum to high temperature stress during reproductive development. *Crop Science* 48(5),
523 1911–1917.

524 Reynolds, M.P., Pierre, C.S., Saad, A.S.I., Vargas, M, Condon, A.G., 2007. Evaluating
525 potential genetic gains in wheat associated with stress-adaptive trait expression in elite

526 genetic resources under drought and heat stress. *Crop Science* 47, 172–189.
527 [10.2135/cropsci2007.10.0022IPBS](https://doi.org/10.2135/cropsci2007.10.0022IPBS)

528 Ristic, Z., Bukovnik, U, Prasad, P.V.V., 2007. Correlation between heat stability of thylakoid
529 membranes and loss of chlorophyll in winter wheat under heat stress. *Crop Science* 47(5),
530 2067–2073.

531 Rodriguez, M., Canales, E., Borrás-Hidalgo, O., 2005. Molecular aspects of abiotic stress in
532 plants. *Biotechnologia Aplicada*.22 (1): 1–10.

533 Rossi, S., Burgess, P., Jespersen, D., Huang, B., 2017. Heat-induced leaf senescence
534 associated with chlorophyll metabolism in bent grass lines differing in heat tolerance. *Crop*
535 *Science*, 57, 169. <http://dx.doi.org/10.2135/cropsci2016.06.0542>

536 Sattar, A., Sher, A., Ijaz, M., Ul-Allah, S., Rizwan, M.S., Hussain, M., Jabran, K, Cheema,
537 M.A., 2020. Terminal drought and heat stress alter physiological and biochemical attributes
538 in flag leaf of bread wheat. *PLoS ONE* 15(5): e0232974.

539 Sharkey, T.D., Zhang, R., 2010. High temperature effects on electron and proton circuits of
540 photosynthesis. *Journal of Integrative Plant Biology* 52(8): 712–722.

541 Todorov, D., Karanov, E., Smith, A.R., Hall, M.A., 2003. Chlorophyllase activity and
542 chlorophyll content in wild and mutant plants of *Arabidopsis thaliana*. *Biologia Plantarum*
543 46(1), 125– 127.

544 Urban, J., Ingwers, M.W., McGuire, M.A., Teskey, R.O., 2017. Increase in leaf temperature
545 opens stomata and decouples net photosynthesis from stomatal conductance in *Pinus taeda*
546 and *Populus deltoides*×*nigra*. *Journal of Experimental Botany* 68(7), 1757–1767.

547 Wang, F.H., Wang, G.X., Li, X.Y., Huang, J.L., Zheng, J.K., 2008. Heredity, physiology and
548 mapping of a chlorophyll content gene of rice (*Oryza sativa* L.). *Journal of Plant Physiology*
549 165(3), 324–330.

550 Van Zanten, M., Voesenek, L.A., Peeters, A.J, Millenaar, F.F., 2009. Hormone- and light-
551 mediated regulation of heat-induced differential petiole growth in *Arabidopsis*. *Plant*
552 *Physiology* 151, 1446–1458. <http://www.plantphysiol.org/cgi/doi/10.1104/pp.109.144386>

553 Yano, T., Aydin, M., Haraguchi, T., 2007. Impact of climate change on irrigation demand
554 and crop growth in a Mediterranean environment of Turkey *Sensors* 7(10), 2297–2315

555 Zhang, Y., Mian, M.A.R., Bouton, J.H., 2006. Recent molecular and genomic studies on
556 stress tolerance of forage and turf grasses. *Crop Science* 46, 497–511.
557 <https://doi.org/10.2135/CROPSCI2004.0572>